

# Changes in the forms of iron and in concentrations of taurine, carnosine, coenzyme Q<sub>10</sub>, and creatine in beef longissimus muscle with cooking and simulated stomach and duodenal digestion

R.W. Purchas<sup>b,\*</sup>, J.R. Busboom<sup>a</sup>, B.H.P. Wilkinson<sup>b</sup>

<sup>a</sup> Department of Animal Sciences, Washington State University, Box 646310, Pullman, WA 99164-6310, USA

<sup>b</sup> Institute of Food, Nutrition and Human Health, Massey University, Private Bag 11 222, Palmerston North, New Zealand

Received 3 March 2006; received in revised form 13 March 2006; accepted 17 March 2006

## Abstract

Longissimus muscle samples from 31 Angus-cross heifers finished on either a high-concentrate feedlot diet or pasture were used to evaluate the effects of cooking (71 °C) on the concentration and forms of iron in the meat, and also on concentrations of the bioactive compounds taurine, carnosine, coenzyme Q<sub>10</sub>, creatine and creatinine. For a sub-sample of 15 the effects of pepsin and pancreatin digestion were assessed. For iron, cooking resulted in some overall loss together with a major change from soluble haem and non-haem iron to the insoluble forms, but solubility was regained to a significant extent following digestion. Total haem iron percentage, however, decreased at each step from an initial 88% to a final 61% of total iron. For the bioactive compounds, cooking led to a reduction in taurine, carnosine, coenzyme Q<sub>10</sub>, and creatine. The effects of digestion on the bioactive compounds varied. No clear differences were shown in the way in which beef from pasture-finished and feedlot-finished cattle responded to cooking and digestion.

© 2006 Elsevier Ltd. All rights reserved.

**Keywords:** Pepsin digestion; Pancreatin; Bile extract; Haem iron; Beef; Cooking

## 1. Introduction

Beef is well recognised as an important dietary source of iron, and particularly of the more bioavailable haem iron (Conrad & Umbreit, 2000; Swain, Tabatabai, & Reddy, 2002). It also contains several compounds with potential bioactive properties including taurine (Huxtable, 1992), carnosine (Decker & Mei, 1996), coenzyme Q<sub>10</sub> (Overvad et al., 1999) and creatine (Burke et al., 2003; Snow & Murphy, 2003). The nature of the bioactive properties of the compounds was outlined by Purchas and Busboom (2005). Whether or not the amounts of these “bioactive”

compounds obtained from typical servings of beef are sufficient to have measurable bioactive effects has not been determined with any certainty. If they are present in useful amounts, then information on their presence in beef would be of value to consumers.

It has been shown that cooking of beef can influence the amount of iron present (Purchas, Simcock, Knight, & Wilkinson, 2003), depending on the time and temperature of cooking, and that large changes in the proportion of iron in the soluble and insoluble forms and in the haem and non-haem forms will occur with most cooking methods (Kristensen & Purslow, 2001; Purchas et al., 2003). Concentrations of the group of bioactive compounds listed above may also change with cooking, either through changes induced by heating, as in the case of the formation of creatinine from creatine (Macy, Naumann, & Bailey,

\* Corresponding author. Tel.: +64 6 350 4336x2536; fax: +64 6 350 5657.

E-mail address: [R.Purchas@massey.ac.nz](mailto:R.Purchas@massey.ac.nz) (R.W. Purchas).

1970), or due to losses of water-soluble compounds such as taurine, carnosine and creatine in cooking juices (Purchas, Rutherford, Pearce, Vather, & Wilkinson, 2004).

The duodenum is the primary site of absorption of iron (Conrad & Umbreit, 2000), so it is of interest to determine the extent to which the forms of iron and the concentrations of the bioactive compounds change during digestion in the stomach and duodenum. The objectives of the research reported here were, first, to measure changes in these concentrations in beef longissimus muscle samples at several steps during the sequential treatments of cooking, in vitro simulated stomach digestion using pepsin plus hydrochloric acid, and simulated duodenal digestion with pancreas extract (pancreatin) plus a bile extract, and, secondly, to determine whether the nature of these changes differed between beef from cattle finished on pasture in New Zealand and beef from similar cattle finished on a high-concentrate ration in the USA.

## 2. Materials and methods

### 2.1. Animals and samples

The 31 Angus-cross heifers from which the longissimus samples were obtained were described by Purchas and Busboom (2005). Briefly, there were 15 heifers aged from 15 to 16 months that had been finished for 98 days on a high-concentrate diet in a feedlot in Washington state, USA (US cattle), and 16 heifers finished on pasture in New Zealand (NZ cattle). The NZ cattle were in two groups, with half that matched the US cattle for age (16–17 months), but were lighter (mean carcass weights of 260 and 322 kg, respectively) (NZAge group), and half that were older (27–28 months), but of a more similar weight to the US cattle (NZWt group). The cattle were slaughtered and dressed in commercial meat plants in the respective countries, and muscle samples (700–900 g), which were taken from the cranial end of *M. longissimus lumborum* on the day following slaughter, were frozen at  $-20^{\circ}\text{C}$  within 36 h postmortem (Purchas & Busboom, 2005).

### 2.2. Cooking and digestion procedures

Muscle samples were thawed at  $1-3^{\circ}\text{C}$  and 30-mm thick steaks were cut for cooking. Two steaks were cut for samples for the NZ cattle and one steak for the US cattle due to the larger cross-section of the latter and the fact that a shorter length of the muscle was available. Steaks were cooked in pairs of approximately the same weight in a clam cooker (Silex Domestic Grill, model 619.80, set at  $200^{\circ}\text{C}$ ) to an internal temperature of  $71^{\circ}\text{C}$  as assessed by Fluke 80PK-5A Type K thermocouple piercing probes attached to a 52 K/J Fluke thermometer, with one probe in each steak. After removal from the grill, steaks were held at room temperature for 10–15 min during which time some cooking juices were released and discarded. The steaks

were then kept at  $1-3^{\circ}\text{C}$  overnight before being cut into strips and homogenised for two periods of approximately 10 s by chopping in a food processor (Magimix Cuisine 5100, using the 300 mL minibowl). Two samples of approximately 70 g were frozen for analysis.

The simulated-digestion protocol based on those of Perez-Llamas, Diepenmaat-Wolters, and Zamora (1997) and Swain et al. (2002), is described below with some of the digestion parameters shown in brackets as means  $\pm$  standard deviations. Approximately 65 g ( $62.4 \pm 6.0$  g) of homogenised cooked sample was added to two volumes of water and the pH adjusted to 2.0 with 6 N HCl ( $5.12 \pm 0.40$  mL) prior to adding 1.3 g pepsin (Sigma P-7000) in 6 mL of 0.1 N HCl. This mix was incubated at  $37^{\circ}\text{C}$  in a water bath with intermittent shaking and with additional 6 N HCl added during the early stages in order to keep the pH close to 2 (total amount added in several lots at 2-min intervals was  $2.14 \pm 0.62$  mL). After 2 h, digestion was stopped by bringing the pH up to 7.0 with 1 M  $\text{NaHCO}_3$  ( $161.3 \pm 20.5$  mL) and two sub-samples of approximately 60 mL ( $63.3 \pm 4.0$  g) were taken and frozen for subsequent analysis. The remainder, or in some cases a sub-sample of 100 mL ( $209.4 \pm 101.2$  mL) was subjected to further digestion by adding 25 mL per 100 mL of a pancreatin plus bile extract solution (0.6 g pancreatin (Sigma P-1750) and 3.75 g bile extract (Sigma B-8631) in 250 mL 0.1 M  $\text{NaHCO}_3$ ) and incubating at  $37^{\circ}\text{C}$  for 1 h in a water bath with intermittent shaking. At the end of this time two samples of approximately 60 mL ( $64.4 \pm 4.9$  g) were taken and the digestion stopped by freezing. Of the two samples taken following cooking, pepsin digestion, and pancreatin digestion, one was used for analysis of the amount and forms of iron directly and the other was freeze dried prior to analysis for taurine, carnosine, coenzyme  $\text{Q}_{10}$ , creatine and creatinine.

### 2.3. Analytical measurements

The proportions of iron as haem iron and non-haem iron in the water-soluble and water-insoluble fractions were assessed as described by Purchas et al. (2003). Briefly, haem iron was assessed using the colorimetric method of Hornsey (1956), and non-haem iron was assayed colorimetrically using the ferrozine method after removal of haem iron by trichloroacetic acid precipitation. Total iron was calculated as the sum of the soluble and insoluble iron.

Taurine, carnosine, coenzyme  $\text{Q}_{10}$ , creatine and creatinine were assayed as described by Purchas et al. (2004). Briefly, taurine and carnosine were quantified using an HPLC system after a buffer extract (67 mM sodium citrate buffer, pH 2.2) had been passed through an ultrafilter with a 5000 MW cutoff. Coenzyme  $\text{Q}_{10}$  was measured in hexane extracts by HPLC. Creatine and creatinine were assayed spectrophotometrically using enzyme-based systems in an auto-analyser (Purchas et al., 2004).

## 2.4. Statistical analysis

Analysis was by a block design used within the GLM Procedure of SAS (SAS Inst. Inc., Cary, NC), with animals as the blocks and the treatments (cooking and digestion) applied within each animal. Thus, the group effect (US cattle vs. NZAge vs. NZWt) was tested against the animal-within-group term, and the between-animal, treatment, and the group-by-treatment interaction effects were tested against the overall error term. Multiple comparisons between groups and between treatments were tested using the least-significant-difference method. For the assessment of cooking effects, data for samples from 31 animals were used, while for the comparison of cooking and digestion effects, there were data for 15 animals.

## 3. Results and discussion

### 3.1. Cooking effects on iron levels

The total weight of steak cooked was lower for the US cattle group (Table 1) mainly because a single steak was cooked for that group while two steaks were cooked for the other two groups, which means that the weights of individual steaks for those groups were lower and consequently the average cooking time was slightly less (Table 1). The absence of any group effect on cooking losses suggests that the differences in steak weights and cooking times were not important sources of variation.

Cooking effects on the level of iron in the longissimus muscle and on the proportion of iron in different forms (Table 2) showed no significant interactions between group and cooking, which indicates that samples from all three groups were affected in a similar way by the cooking treatment. The group effect shown for uncooked meat was

Table 1

Mean steak weights, cooking losses and cooking times for longissimus muscle samples from the three groups of Angus-cross heifers

	Steak weight (g)	Cooking loss (%)	Cooking time (min)
Group			
US cattle ( <i>n</i> = 15)	223.2a	32.4	6.64b
NZAge ( <i>n</i> = 8)	384.6b	31.6	5.05a
NZWt ( <i>n</i> = 8)	387.8b	31.1	5.79a
Group effect <sup>A</sup>	***	ns	**
<i>R</i> <sup>2</sup> (%), RSD <sup>B</sup>	82, 40.6	8, 2.04	38, 0.90

<sup>A</sup> \*\*\*, *P* < 0.001; \*\*, *P* < 0.01; ns, *P* > 0.10.

<sup>B</sup> Measures of goodness of fit of the model are given by coefficients of determination [*R*<sup>2</sup> (%)] and residual standard deviations (RSD). Group means within a column do not differ significantly (i.e., *P* > 0.05) if they have a common letter or no letter.

retained for cooked meat with the NZWt group having higher levels of iron than the other two groups, and a lower proportion of soluble non-haem iron. The proportion of insoluble haem iron was higher for that group. The effects of cooking were similar to those that have been reported previously (Kristensen & Purslow, 2001; Purchas et al., 2003; Schricker & Miller, 1983) with a marked shift from soluble to insoluble haem iron percentage, a decrease in soluble non-haem, and a big increase in insoluble non-haem iron. The last of these changes reflects the overall reduction in total percentage of iron in the haem form. The decrease in total iron was probably due to iron losses in the cooking juices (Purchas et al., 2003).

### 3.2. Digestion effects on iron levels

Analyses of changes taking place during simulated stomach and duodenal digestion for muscle samples from 15 animals (Table 3) again revealed no significant group by treatment interactions. Group differences were similar

Table 2

Levels of iron and the proportions of iron in the soluble and insoluble haem and non-haem forms in the longissimus muscle of 31 heifers with and without cooking

	Total iron (μg g <sup>-1</sup> )	Soluble iron (%)		Insoluble iron (%)		Total haem iron (%)
		Haem	Non-haem	Haem	Non-haem	
Group (means across uncooked and cooked samples)						
US cattle ( <i>n</i> = 15)	13.50a	40.3	5.5b	41.7a	12.5	82.0
NZAge ( <i>n</i> = 8)	14.85a	39.8	5.1b	40.5a	14.6	80.3
NZWt ( <i>n</i> = 8)	18.80b	38.0	3.8a	44.7b	13.4	82.8
Group effect <sup>A</sup>	***	+	**	*	+	ns
Cooking treatment						
Uncooked ( <i>n</i> = 31)	17.45	75.3	5.4	12.4	6.9	87.7
Cooked ( <i>n</i> = 31)	13.99	3.4	4.2	72.2	20.1	75.6
Cooking effect <sup>A</sup>	**	***	**	***	***	***
<i>R</i> <sup>2</sup> (%), RSD <sup>B</sup>	96, 1.08	99, 3.75	61, 1.72	99, 5.12	92, 3.08	88, 3.59

Interactions between group and cooking treatment were not significant (*P* > 0.05). Total iron concentrations are relative to uncooked weight and all other values are as percentages of total iron.

<sup>A</sup> \*\*\*, *P* < 0.001; \*\*, *P* < 0.01; \*, *P* < 0.05; +, *P* < 0.10; ns, *P* > 0.10.

<sup>B</sup> Measures of goodness of fit of the model are given by coefficients of determination [*R*<sup>2</sup> (%)] and residual standard deviations (RSD). Group means within a column do not differ significantly (i.e., *P* > 0.05) if they have a common letter or no letter.

Table 3

Levels of iron and the proportions of iron in the soluble and insoluble haem and non-haem forms in the longissimus muscle from 15 heifers following cooking, and simulated stomach (pepsin/HCl treatment) and duodenal (pancreatin/bile treatment) digestion

	Total iron ( $\mu\text{g g}^{-1}$ )	Soluble iron (%)		Insoluble iron (%)		Total haem iron (%)
		Haem	Non-haem	Haem	Non-haem	
Group (means across all treatments)						
US cattle ( $n = 7$ )	13.79a	25.8	16.2	46.4	11.6a	72.2
NZAge ( $n = 4$ )	16.61ab	25.3	14.0	48.2	12.4b	73.6
NZWt ( $n = 4$ )	17.42b	25.0	13.5	49.6	11.8a	74.7
Group effect <sup>A</sup>	*	ns	+	ns	*	ns
Treatment						
Uncooked	16.90c	75.5c	5.2a	12.2a	7.1a	87.7d
Cooked	13.65a	2.9a	4.6a	74.2d	18.3d	77.1c
Pepsin/HCl	15.51b	2.7a	18.6b	65.7c	13.0c	68.4b
Pancreatin/bile	17.69c	20.6b	29.9c	40.1b	9.4b	60.7a
Treatment effect <sup>A</sup>	***	***	***	***	***	***
$R^2$ (%), RSD <sup>B</sup>	88, 1.40	99, 3.4	95, 3.1	98, 4.8	90, 1.8	95, 3.3

Interactions between group and treatment were not significant ( $P > 0.05$ ). Total iron concentrations are relative to uncooked weight and all other values are as percentages of total iron.

<sup>A</sup> \*\*\*,  $P < 0.001$ ; \*\*,  $P < 0.01$ ; \*,  $P < 0.05$ ; +,  $P < 0.10$ ; ns,  $P > 0.10$ .

<sup>B</sup> Measures of goodness of fit of the model are given by coefficients of determination [ $R^2$  (%)] and residual standard deviations (RSD). Group or treatment means within a column do not differ significantly (i.e.,  $P > 0.05$ ) if they have a common letter or no letters.

to those shown for the larger group of animals (Table 2), with the levels of significance lower due to the smaller numbers.

Changes with digestion shown in Table 3 and Fig. 1 for total iron are difficult to explain. The reduction with cooking was probably due to losses in cooking juices, but the increase following pepsin digestion, and the further increase following pancreatin digestion were not expected, which suggests that some of the iron may not have been detected in any of the four fractions following cooking

and pepsin treatment. A possible explanation is that the trichloroacetic acid precipitation step that was used to remove the haem iron prior to assaying for non-haem iron, also removed some non-haem iron for the cooked and pepsin-treated samples. If this was the case, the percentages of non-haem fractions, and therefore total iron as well, following cooking and pepsin/HCl treatment (Table 3) may be slightly underestimated.

With the two-step digestion process, the changes in the proportions of iron in the four fractions relative to the cooked samples included the following:

- For soluble haem iron, pepsin digestion had no effect, but pancreatin/bile treatment increased this percentage over seven-fold, due to a conversion from the insoluble form of haem iron, but the soluble haem iron percentage was still less than one third that of the uncooked samples.
- For soluble non-haem iron, pepsin treatment brought about a significant four-fold increase in the percentage, which was increased a further 61% by pancreatin, to give a final value that was more than five times the cooked value or the uncooked value. This was presumably due to a solubilisation of haem-containing proteins or peptides that had been denatured by cooking.
- For insoluble haem iron, both pepsin and pancreatin treatments led to a reduction in the percentage, which is consistent with the increase in soluble haem iron, and with previous reports of the effect of proteolytic digestion (Seth, Diaz, & Mahoney, 1999).
- For insoluble non-haem iron, the pattern was similar to insoluble haem iron with pepsin and pancreatin leading to lower percentages, but the final percentage was still higher than for uncooked meat.

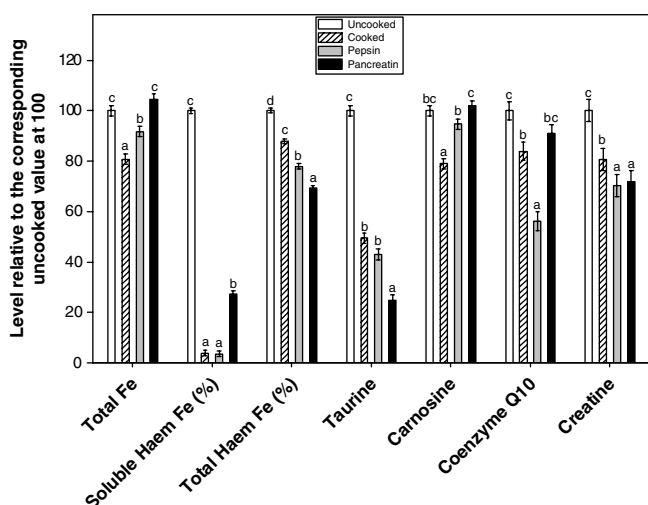


Fig. 1. Means ( $\pm$ SE) for levels of several items within the longissimus muscle from 15 Angus-cross heifers showing the accumulative effects of cooking, pepsin digestion, and pancreatin digestion. Levels are all given relative to the corresponding uncooked value which is set at 100 to permit use of a common scale. Bars within the groups of four without a common letter above them differ significantly ( $P < 0.05$ ).

- For total haem iron, the percentage decreased at both steps indicating that the reduction in insoluble haem iron reflected both a conversion to the soluble form as well as some conversion to the non-haem form.

Overall the digestion changes included some that would be expected to improve iron bioavailability (increased percentages of soluble haem iron and soluble non-haem iron) and some that are likely decrease that bioavailability (decreased percentage of total haem iron) (Conrad & Umbreit, 2000).

### 3.3. Cooking effects on items other than iron

The cooking treatment brought about a reduction in all the compounds shown in Table 4 except for the increase in creatinine levels, which is likely to be attributable to heat-induced conversion of creatine to creatinine (Macy et al., 1970; Purchas et al., 2004). It is not possible to determine the extent to which the reductions with cooking were due to losses in cooking juices or to changes in the compounds. The decrease in creatine concentration with cooking (by  $0.56 \text{ mmol } 100 \text{ g}^{-1}$ ) was appreciably greater than the increase in creatinine ( $0.33 \text{ mmol } 100 \text{ g}^{-1}$ ), indicating that losses could not all be due to a conversion of creatine to creatinine. A similar pattern was reported for lamb muscles cooked for 90 min at  $70^\circ\text{C}$  by Purchas et al. (2004), but in that case the discrepancy between creatine loss and creatinine accumulation was greater.

Significant interactions between group and cooking existed for carnosine and coenzyme  $\text{Q}_{10}$  (Table 4). For carnosine this reflected the fact that a significant group effect was shown for uncooked meat only, but the decrease in carnosine levels following cooking as shown in Table 4 was similar within each group. For coenzyme  $\text{Q}_{10}$  the group effect was the same for cooked and uncooked meat with a lower level in meat from US cattle, but the cooking

effect was more significant for the NZWt group than the other two groups.

### 3.4. Digestion effects on items other than iron

Digestion of cooked samples of meat from a sub-sample of animals in a two-step process brought about the changes outlined below (Table 5) relative to the cooked samples (all values within a column in Table 5 are directly comparable as they are expressed relative to  $100 \text{ g}$  of the initial uncooked meat):

- Taurine levels were unaffected by pepsin treatment, but were significantly lower following digestion with pancreatin. Running a taurine standard through the digestion process indicated that the lower levels were not due to the breakdown of taurine, but it may indicate that some of the taurine became inaccessible in some way. Taurine standard did not change significantly when subjected to the same digestion process, but it is possible that components from the digested meat may have restricted the accessibility of the taurine as the aqueous buffer extract was passed through a  $5000 \text{ kDa}$  MW filter (Purchas et al., 2004).
- Carnosine levels increased by 20% following pepsin digestion, and by a further 8% following pancreatin digestion. Again, the explanation for these increases is not known, but it is possible that protein breakdown that occurred during digestion resulted in the formation of carnosine-like dipeptides. The increases were not due to carnosine in the digestion reagents as the values have been adjusted for reagent blanks that were run through the system.
- Coenzyme  $\text{Q}_{10}$  levels decreased following pepsin digestion, but increased to values not significantly different from the cooked sample levels following pancreatin digestion.

Table 4

Concentrations relative to uncooked weight of taurine, carnosine, coenzyme  $\text{Q}_{10}$ , creatine and creatinine in the uncooked and cooked longissimus muscle samples from 31 heifers in three groups

	Taurine ( $\text{mg } 100 \text{ g}^{-1}$ )	Carnosine ( $\text{mg } 100 \text{ g}^{-1}$ )	Coenzyme $\text{Q}_{10}$ ( $\text{mg } 100 \text{ g}^{-1}$ )	Creatine ( $\text{mg } 100 \text{ g}^{-1}$ )	Creatinine ( $\text{mg } 100 \text{ g}^{-1}$ )
Group (means across uncooked and cooked samples)					
US cattle ( $n = 15$ )	37.7	351.0a	0.94a	351.6b	25.48
NZAge ( $n = 8$ )	37.4	396.0b	1.50b	330.5a	21.51
NZWt ( $n = 8$ )	38.2	383.0ab	1.53b	358.9b	27.11
Group effect <sup>A</sup>	ns	**	***	**	ns
Cooking treatment					
Uncooked	51.0	433.0	1.44	383.5	6.15
Cooked	24.5	321.0	1.21	310.6	43.25
Cooking effect <sup>A</sup>	***	***	***	***	***
Group $\times$ cooking interaction <sup>A</sup>	ns	***	**	ns	ns
$R^2$ (%), RSD <sup>B</sup>	95, 4.7	83, 46.2	91, 0.18	95, 14.6	93, 7.89

<sup>A</sup> \*\*\*,  $P < 0.001$ ; \*\*,  $P < 0.01$ ; \*,  $P < 0.05$ ; +,  $P < 0.10$ ; ns,  $P > 0.10$ .

<sup>B</sup> Measures of goodness of fit of the model are given by coefficients of determination [ $R^2$  (%)] and residual standard deviations (RSD). Group means within a column do not differ significantly (i.e.,  $P > 0.05$ ) if they have a common letter or no letter.

Table 5

Concentrations relative to uncooked weight of taurine, carnosine, coenzyme Q<sub>10</sub>, creatine and creatinine in the longissimus muscles of 15 heifers following cooking, and simulated stomach (pepsin/HCl treatment) and duodenal (pancreatin/bile treatment) digestion

	Taurine (mg 100 g <sup>-1</sup> )	Carnosine (mg 100 g <sup>-1</sup> )	Coenzyme Q <sub>10</sub> (mg 100 g <sup>-1</sup> )	Creatine (mg 100 g <sup>-1</sup> )	Creatinine (mg 100 g <sup>-1</sup> )
Group (means across all treatments)					
US cattle ( <i>n</i> = 7)	28.3	363.2a	0.87a	310.8	37.04b
NZAge ( <i>n</i> = 4)	28.9	427.8b	1.52b	303.0	30.27a
NZWt ( <i>n</i> = 4)	23.6	363.2a	1.46b	318.6	39.36b
Group effect <sup>A</sup>	+	**	***	ns	*
Treatment					
Uncooked	49.6c	409.6bc	1.55c	384.9c	6.05a
Cooked	24.3b	323.5a	1.30b	311.1b	43.75b
Pepsin/HCl	21.3b	387.9b	0.87a	270.5a	44.05b
Pancreatin	12.4a	418.0c	1.41bc	276.7a	48.38c
Treatment effect <sup>A</sup>	***	***	***	***	***
Group × Treatment interaction <sup>A</sup>	ns	*	ns	ns	ns
R <sup>2</sup> (%), RSD <sup>B</sup>	96, 4.0	82, 31.9	86, 0.22	93, 16.7	96, 5.03

<sup>A</sup> \*\*\*, *P* < 0.001; \*\*, *P* < 0.01; \*, *P* < 0.05; ns, *P* > 0.05.

<sup>B</sup> Measures of goodness of fit of the model are given by coefficients of determination [*R*<sup>2</sup> (%)] and residual standard deviations (RSD). Group or treatment means within a column do not differ significantly (i.e., *P* > 0.05) if they have a common letter or no letter.

- Creatine levels decreased by 40.6 mg 100 g<sup>-1</sup> (13%) with pepsin digestion, but showed no further change with pancreatin digestion. The decrease with pepsin was not accompanied by an increase in creatinine, which suggests that some other form of change occurred.
- Creatinine levels were unaffected by pepsin, but increased slightly with pancreatin treatment.

The only significant interaction between group and treatment for the analyses shown in Table 5 was for carnosine where the ranking of the four treatments was the same for each of the groups except that, for the NZWt group, levels were lower following pepsin digestion, while for the other two groups they were higher.

#### 4. Conclusion

Some of the undesirable changes in the solubility and form of iron in beef brought about by cooking were reversed to some extent by digestion processes. The ways in which the iron fractions in beef responded to cooking and digestion treatments were similar for beef from feedlot-finished and pasture-finished cattle. Significant changes were found to take place in the levels of taurine, carnosine, coenzyme Q<sub>10</sub>, creatine, and creatinine with cooking and digestion, but the nature of these changes were similar for beef from cattle finished in different production systems.

#### Acknowledgments

This research was carried out under contract to Meat New Zealand. The technical contributions by Dr. Philip Pearce, Ms. Maggie Zou, Ms. Leiza Turnbull and Ms. Rosheila Vather are gratefully acknowledged.

#### References

- Burke, D. G., Chilibeck, P. D., Parise, G., Candow, D. G., Mahoney, D., & Tarnopolsky, M. (2003). Effect of creatine and weight training on muscle creatine and performance in vegetarians. *Medicine and Science in Sports and Exercise*, 35, 1946–1955.
- Conrad, M. E., & Umbreit, J. N. (2000). Iron absorption and transport – an update. *American Journal of Hematology*, 64, 287–298.
- Decker, E. A., & Mei, L. (1996). Antioxidant mechanisms and applications in muscle foods. *Reciprocal Meat conference Proceedings*, 49, 64–72.
- Hornsey, H. C. (1956). The colour of cooked cured pork. 1. Estimation of the nitric oxide-haem pigments. *Journal of the Science of Food and Agriculture*, 7, 534–540.
- Huxtable, R. J. (1992). Physiological actions of taurine. *Physiological Reviews*, 72, 101–163.
- Kristensen, L., & Purslow, P. P. (2001). The effect of processing temperature and addition of mono- and di-valent salts on the heme- and nonheme-iron ratio in meat. *Food Chemistry*, 73, 433–439.
- Macy, R. L., Naumann, H. D., & Bailey, M. E. (1970). Water-soluble flavour and odor precursors of meat. 5. Influence of heating on acid-extractable non-nucleotide chemical constituents of beef, lamb and pork. *Journal of Food Science*, 35, 83–87.
- Overvad, K., Diamant, B., Holm, L., Holmer, G., Mortensen, S. A., & Stender, S. (1999). Review: Coenzyme Q<sub>10</sub> in health and disease. *European Journal of Clinical Nutrition*, 53, 764–770.
- Perez-Llamas, F., Diepenmaat-Wolters, M. G. E., & Zamora, S. (1997). Influence of different types of protein on in vitro availability of intrinsic and extrinsic iron and zinc. *Journal of the Science of Food and Agriculture*, 75, 303–311.
- Purchas, R. W., & Busboom, J. R. (2005). The effect of production system and age on levels of iron, taurine, carnosine, coenzyme Q<sub>10</sub>, and creatine in beef muscles and liver. *Meat Science*, 70, 589–596.
- Purchas, R. W., Rutherford, S. M., Pearce, P. D., Vather, R., & Wilkinson, B. H. P. (2004). Concentrations in beef and lamb of taurine, carnosine, coenzyme Q<sub>10</sub>, and creatine. *Meat Science*, 66, 629–637.
- Purchas, R. W., Simcock, D. C., Knight, T. W., & Wilkinson, B. H. P. (2003). Variation in the form of iron in beef and lamb meat and losses of iron during cooking and storage. *International Journal of Food Science and Technology*, 38, 827–837.

- Schricker, B. R., & Miller, D. D. (1983). Effects of cooking and chemical treatment on heme and nonheme iron in meat. *Journal of Food Science*, 48, 1340–1343.
- Seth, A., Diaz, M., & Mahoney, R. R. (1999). Iron solubilisation by chicken muscle protein digests. *Journal of the Science of Food and Agriculture*, 79, 1958–1963.
- Snow, R. J., & Murphy, R. M. (2003). Factors influencing creatine loading into human skeletal muscle. *Exercise and Sports Science Review*, 31, 154–158.
- Swain, J. H., Tabatabai, L. B., & Reddy, M. B. (2002). Histidine content of low-molecular-weight beef proteins influences nonheme iron bioavailability in Caco-2 cells. *Journal of Nutrition*, 132, 245–251.